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### REACTIONS OF DROSOPHILA TO 2537Å RADIATION

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Many insects react to at least the longer wave-lengths of ultraviolet light as though they see them quite clearly. Kühn and Pohl (1921, Naturwiss., IX, p. 738) trained the honey-bee, Apis, to come to certain ultraviolet wave-lengths for food, and Lutz (1923, Amer. Mus. Novitates, No. 641) showed that another bee, Trigona, distinguished between ultraviolet patterns, one of which marked the nest-site.

Bertholf has published several very careful pieces of work on this subject. One (1931, Jour. Agric. Research, XLIII, p. 703) concerned Apis. In it he gives a curve of the "stimulative efficiency" of light from a "Uviare" lamp giving radiation from about 2800 to about 7000 Å. This curve has a low peak near 5500 and a high one in the ultraviolet at 3650 Å. Light from the mercury-vapor arc was dispersed by a spectroscope and brought to focus on a piece of ground quartz. "Visible light" from an incandescent electric lamp illuminated a piece of ground glass, the intensity of illumination being controlled by the experimenter. Bees, put into an otherwise dark box, tended to go to whichever of these illuminated windows seemed brightest to them. An intensity of white light was found such that the bees went about equally to it and to the window of ground quartz illuminated by that one of the spectral lines which was being investigated. Knowing the relative intensity of the white light and the energy of the spectral band (after it had passed through the ground quartz?), Bertholf obtained a coefficient of the "relative stimulative efficiency" of the spectral band. However, the energy from the source used dropped off at 2800 Å to a quantity too small to be measured by a thermopile and galvanometer. It is not strange, therefore, that 2800 Å did not show any relative stimulative efficiency. This leaves open the question as to how far into the ultraviolet this bee can see.

In a subsequent paper (1932, Zeit. f. vergl. Physiologie, XVIII, p. 32) Bertholf used the same experimental method with a fly, *Drosophila melanogaster*. In this case the source of ultraviolet had slight but measurable energy in radiation as short as 2380 Å. He states that the energy at 2350 Å was "too small to measure" and that the relative

stimulative effect there was "very slight if any." In view of the difficulty of transmitting through air radiation of shorter wave-lengths than about 2500 Å, it is indeed surprising that the flies showed any response. Going up the scale toward longer waves, a maximum efficiency was found at about 3650 Å. There was, however, an indication of a minor peak at about 2540, probably the band near 2537 Å, the one which we used in the experiments described in this paper. It is possible that this peak was due, at least in part, to the fact that his source of ultraviolet had a "relative energy" of 7.3 there as compared with 0.9 at 2480 and with 1.8 at 2800 Å. It is also possible that, were we able to transmit through air a reasonable amount of radiation shorter than 2300 Å we might find that insects react to it as well.

Our experiments with the same fly, *Drosophila melanogaster*, dealt solely with the 2537 line, using a different experimental method. They confirm Bertholf's findings as to the fact that the fly reacts to this radiation. The apparatus used was as follows:

A cold quartz-mercury arc was focused by a quartz condensing lens on the slit of a small quartz spectrograph (Hilger type E 37). The rear aperture of the spectrograph was covered with a black cardboard, in which a narrow slit passed the 2537 line but no others.

The specimens were put into a glass tube 5 cm. long with an internal diameter of about 4 mm. Each end of the tube was fitted with removable quartz windows approximately 2 mm. thick. A black cardboard mask was placed between each window and the tube. Each mask had a hole fitting exactly the bore of the tube, and the outer edges of each mask extended well beyond the outside of the tube, thus preventing light from entering the tube except at the ends of the bore.

The tube was mounted on a leveling table back of the spectrograph. Adjustment to 2537 was checked at both ends of the tube by means of a fluorescent glass. The 2537 line was made sufficiently high to fill the tube vertically, but it was barely half a millimeter wide where it entered the left-hand end of the tube and only about 3 mm. wide at the other end. Owing to the small size of *Drosophila*, it was easily possible for such a fly to rest in parts of the tube with its eyes entirely out of the 2537 radiation.

Long-wave radiation, visible to man, was obtained from an incandescent electric light focused through an f 4.5 camera lens. In the work reported here a 100-watt "spot-light" was used. The filament was focused about 10 cm. in front of the right-hand end of the specimen tube so that the light entered the tube as a diverging beam, completely filling it. Stray light was largely prevented by the housing of the lamp.

Threads, one centimeter apart, were tied around the tube, marking off five divisions that are referred to here as I (the centimeter nearest the source of the short-wave radiation), II, III, IV, and V (the centimeter nearest the source of the long-wave radiation).

In order to observe the actions of the flies at all times, a small microscope lamp was hung about 50 cms. above the specimen tube. window of this lamp was ground glass and for this work was covered with a red filter of such a density that a fly could barely be seen in the tube when other lights were off. The ventilation holes of this lamp permitted some light to escape toward the ceiling of the room. Also, since the mercury arc was not completely enclosed, stray light from it was diffused about the room when the arc was running. Scattered light from these sources was very weak and generally distributed. However, the I end of the specimen tube was close to the black screen of the spectrograph, and the V end was pointed toward the open room. The slight tendency of the flies to favor the V end may have been due to this weak scattered light in the room. If so, it would tend to counteract any "attractive" effect the 2537 radiation might have had. An attempt has been made in the analysis of the results to neutralize this possible source of error.

Reflections within the tube and fluorescence of it should also be kept in mind. The long-wave radiation, filling the tube, was in part reflected from its sides at a slight angle. Some was reflected directly backward from the quartz window at the I end. On the other hand, the 2537 beam did not nearly fill the tube until close to the V end, where some was probably reflected backward by the quartz window there. Where the 2537 beam struck the glass tube, it set up an exceedingly faint fluorescence noticeable to us only when the red light was off, and our eyes were completely dark-adapted. It was slightly more intense but very limited in area at the I end; more diffuse at the V end. If these reflections and fluorescence had any effect on the actions of the flies, they probably tended to decrease the apparent effectiveness of the 2537 beam and to increase the apparent effectiveness of the light visible to us.

A record was kept of the division of the tube in which the fly was at each second of the experiment. This was done by means of a typewriter and a metronome beating seconds. Five typewriter keys in a row were taken to represent the five divisions of the tube. At each beat of the metronome that key which corresponded to the division occupied by the fly was pressed.

Five conditions of illumination were considered: "Dr," darkness except for the red light and such other light as was scattered from its ventilation holes; "Dra," both the red light and the mercury are going but the ultraviolet components of the beam coming through the condenser lens cut off from the spectrograph by two sheets of glass (total thickness, 4 mm.) placed between the lens and the slit; "Lr," long-wave light entering the V end of the tube and the red light on; "Sra," the 2537 beam entering the I end of the tube and the red light and, of course, the arc on; and "SLra," a combination of Lr and Sra.

In the fifteen experiments summarized in Table I, each fly was subjected for one minute to each of these conditions of illumination in the sequence, Dr, Lr, Dr, Sra, Dra, SLra, the cycle being run five times in as quick succession as possible. In this way Lr was checked by the Dr both before and after it, while Dra served as a check on the immediately preceding Sra and the immediately following SLra.

The flies used were bred and kept in the dark-room. The transferring of a fly from the breeding jar to the observation tube was done quickly in illumination from an incandescent lamp, and then the tube was left in Dr for several minutes before starting the experiment. This delay gave the fly time to quiet down somewhat and also to regain what dark-adaptedness it might have lost during the transfer. Usually the flies kept up a fair state of activity in the tube for several hours—much longer than was required—but a few individuals were inclined to remain quietly in one place, particularly at one end or the other of the tube. Several very inactive flies were discarded entirely, but otherwise those noted are not selected individuals.

#### BEHAVIOR IN "DARKNESS"

If the position of a fly in the tube were a purely random matter, we might expect an equal number of records for each division. However, owing to the presence of a window at each end, there was a greater area over which the fly could walk in I and in V than in either II, III, or IV. In accord with this, it will be noticed that the flies were in each of the end divisions more than in any of the intermediate ones. The middle division, III, was favored by the fact that the flies must go through it when moving from the two divisions at either end to the other.

However, I plus II would be expected to equal IV plus V unless there was something particularly attractive at one end or the other. Combining the records of all fifteen flies, out of the 8940 seconds in Dr the flies spent 3414 in I and II and 3719 in IV and V. This is in the proportion of 0.48 to 0.52. The difference from the expected (on the basis of chance) proportion of 0.50 each is not very significant but should be kept in mind. In the case of Dra the proportions are 1621 to 2047 or 0.44 to 0.56, clearly significant differences from 0.50 each.

Since everything about the tube itself was the same during the Dr and the Dra runs for individual flies, it is probable that the slight favoring of the IV+V end of the tube when only the red light was on and the stronger favoring when the arc also was burning are to be explained as the effect of stray light from these sources being reflected from the walls of the room into that end of the tube. As is to be expected on the basis of chance and because of variability in the sensitiveness of flies, this bias in favor of the IV+V end of the tube is not apparent in the case of every individual fly. So far as it existed it would increase the apparent effectiveness of the long-wave radiation and decrease the apparent effectiveness of the 2537 beam.

#### BEHAVIOR IN LONG-WAVE RADIATION

The "visible light" as it entered the V end of the tube had an intensity of 120 foot candles. The fly could easily be seen in this illumination, but, for the sake of uniformity, the red light (with a little scattered light from the lamp's ventilation holes) was kept on. Table I gives the record for 4500 seconds. This is reduced in Table II to terms of the percentages of time spent in I+II: III: IV+V, giving 30, 12, and 58. The corresponding percentages during Dr were 38, 20, and 42. Subtracting the latter from the former shows the shift caused by long-wave radiation entering the V end of the tube. It gives —8 for I+II, —8 for III, and +16 for IV+V. The increase in the two-fifths of the tube nearest the source of long-wave light is clearly significant. That normal Drosophila melanogaster goes toward light visible to us is not news. The data are given here for the sake of comparison with the fly's reactions to 2537Å.

## Behavior in 2537Å

The beam of 2537Å radiation as it struck the outer face of the quartz window on the I end of the tube had an intensity of 8 microwatts per sq. cm. This radiation is well beyond the ultraviolet region usually considered in biological work. Significantly shorter wave-lengths than this (until X-rays¹ are reached) are so fully absorbed by air that they can be well transmitted only in a vacuum. In fact, even 2537 is absent from sunlight at moderate altitudes. Since it is probable that atmos-

<sup>&</sup>lt;sup>1</sup>So far as we know, the conclusion of Axenfold (1896, Centralblatt Physiologie, X, p. 436) that photopositive insects react to X-rays has not been disproved. This should be retested, carefully guarding against possible errors caused by fluorescence.

pheric conditions in past geologic ages were even less favorable for its transmisson, there is no reason to believe that insects have ever encountered it in nature.

Table I shows that when the 2537 line entered the tube (Sra) the flies spent 2920 out of 4500 seconds in the two centimeters at that end of the tube (I+II). This is 65 per cent of the time and is to be contrasted with Dra during which all of the conditions were the same except that 4 mm. of plain glass prevented the 2537 radiation from entering the spectrograph. In Dra the flies spent 1621 out of 4500 seconds in the two centimeters at the spectrograph end of the tube. This is only 36 per cent of the time. The difference between Sra and Dra is 29 per cent, making it quite clear that the 2537 radiation has a marked effect on the flies' behavior.

This effect of 2537 is all the more striking when it is noted that, with the possible exception of fly 18;3 (see Table III and the accompanying footnote), no individual fly responded with a difference in I+II of less than 11 between Sra and Dra, while four of them had a difference between Lr and Dr (proportionate time spent in IV+V) of less than 11. The average difference between IV+V in Lr and the same in Dr was only 16 as compared with 29 for Sra and Dra. Furthermore, as was pointed out before, the beam of 2537 covered only about an eighth of the horizontal diameter of the tube at the spectrograph end, leaving considerable space dark. Had we been able to fill the tube completely with 2537 light, the effect would probably have been even more evident.

#### LONG-WAVE IN COMPETITION WITH 2537 RADIATION

In this part of the experiment (SLra) long-wave light entered the V end of the tube and 2537 entered the I end. The 4500 seconds were divided in the proportions of 47:11:42 among I+II, III, and IV+V, respectively (see Table IV). The difference in proportions between the two ends is only 5. However, this does not take into account the favoring of the V end in Dra. If we subtract from the proportions in SLra the proportions in Dra, we get +11, -8, and -3. While these numbers are too small to permit a very definite statement, the indications are that, as used in these experiments, the 2537 beam of about 8 microwatts per sq. cm. was sufficiently "attractive" to this photopositive insect to more than balance the "attractiveness" of the 120 foot candles white light visible to us.

Since the intensity of the long-wave radiation, measured by a calibrated photronic cell and microammeter, is given in foot candle power, 1934]

and the intensity of the 2537 radiation, measured by a thermopile, is given in microwatts per sq. cm., they are not comparable. Where "white light" is used, the statement of its intensity in terms of total energy does not seem entirely justified because, even if infrared is filtered out, there remain in white light a large number of bands which do not have equal effectiveness per unit of energy, and their relative effectiveness, comparing one with another, varies with the intensity. The only certain way would seem to be the using of monochromatic light in the long-wave as well as in the short-wave region. This was not attempted in the present work.

So far as we have been able to recognize them, all uncertain factors, such as the number of times the flies got out of the narrow beam of 2537 radiation and the effect of reflections within the tube, tended to decrease the apparent effectiveness of the 2537 radiation. Therefore, we believe that its real effectiveness must be even greater than our data indicate.

### Do the Flies See 2537Å Radiation?

Granted that *Drosophila* behaves as though it sees the 2537 beam, there is still a possibility that it does not actually see such light directly but that the 2537 radiation sets up a fluorescence in the fly's eye and that the fly reacts to the presence of this fluorescence. Such a fluorescence, if present, might have a wave-length or wave-lengths greater than 4000 and, so, be in our visual range or it might be in the near ultraviolet. In the latter case the fly would still be directly seeing ultraviolet but not such an extremely short wave-length.

Before radiation can reach the sensitive parts of an insect's eye it must pass through a hard outer layer, the cornea. Unfortunately, mounting the cornea of a *Drosophila* eye so that a spectrogram could be made of the radiation transmitted by it did not seem feasible. We did, however, make spectrograms of the radiation transmitted by the cornea of a large flesh-fly (*Sarcophaga*) and by that of the hive-bee (*Apis*). Figure 1 shows that not only does the cornea of each of these very dissimilar insects transmit all of the ultraviolet bands of the source used but that it is distinctly more transparent to the extremely short wave (2537) than is even cellophane approximately 0.025 mm. thick.<sup>1</sup>

<sup>&#</sup>x27;The unpigmented cuticula of the third abdominal tergum of the bee was found to transmit rather fully the near ultraviolet, but only very faintly the 2537. The pigmented cuticula of the thorax of the Sarcophaga did not transmit either. For further and more detailed information concerning fluorescence in insect eyes and the permeability of insect exoskeletons to ultraviolet light, see, for example, Merkers, 1929, Zool. Jahrb., Abt. Allg. Zool. u. Physiol., XLVI, p. 483. It is thinkable that reactions to extreme ultraviolet are due to sensations more nearly akin to "feeling" than to "seeing." This can scarcely be true of the near ultraviolet where Trigona distinguished detailed patterns. We did not try blinded Drosophila, but the regularity of Bertholf's curve and the fact that there was no change in the type of the fly's reactions when offered 2537 indicate that we are really dealing with vision. A wingless race, notably unreacting to light, did not, so far as our experiments went, react to 2537.

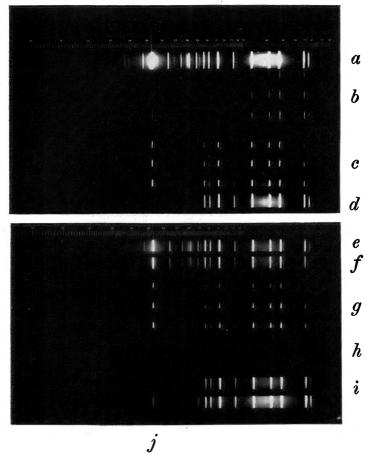


Fig. 1.—Spectrograms of: a, the arc direct, 15 secs.; b, the arc through the exoskeleton of the dorsum of the abdomen of A pis, 15, 30, and 60 secs.; c, the arc through the cornea of A pis, 15, 30, and 60 secs.; d, the arc through cellophane, 15 secs.; e, the arc direct, 15 secs; f, the arc through quartz, 15 secs.; g, the arc through the cornea of Sarcophaga, 15, 30, and 60 secs.; h, the arc through the exoskeleton of the dorsum of the thorax of Sarcophaga, 15 and 30 secs. (no transmission); i, the arc through cellophane, 15 and 60 secs. The 2537Å line is seen at j.

This being true, it follows that not only does the 2537 radiation reach the inner elements of the eye but also that it probably causes no fluorescence on the cornea. Fluorescence can not be set up unless energy is absorbed, and our plates show neither such absorption nor fluorescent bands.

Even so, it might be possible that fluorescence is produced within the eye, and that this fluorescence, rather than direct 2537 radiation, affects the optic nerves. When we let the 2537 beam fall on a mass of crushed Drosophila eyes we could detect no fluorescence. While this does not disprove the presence of fluorescence, it would seem to indicate that, if there be any in our visual range, it is very faint—apparently much too faint to more than counterbalance the long-wave light from the incandescent lamp in the SLra experiments. If fluorescence be present but be of ultraviolet wave-length, we come back to the conclusion that these insects see ultraviolet.

It was thought that possibly some evidence on this subject might be obtained by experiments in light-fatigue. If we can not fatigue the fly's eves to 2537 without affecting its reactions to wave-lengths greater than 4000 and vice versa, the explanation of the failure might be either (1) that the fly's eyes can not readily be fatigued in the wave-length tried or (2) that the fly does not see 2537 directly but as a fluorescence in the long waves, and, so, in attempting to fatigue it to 2537 we are really fatiguing it in light of longer waves. On the other hand, if it can be done, it would indicate that the fly sees 2537 directly, although there is a possible objection that the phenomenon is still one of fluorescence but that there is a fluorescence in a narrow band of wave-lengths and that we are fatiguing to that narrow band in contrast to the wide range of wave-lengths in light visible to us.

In experiment 20:1 a female was subjected for nearly three-quarters of an hour to a constant counterillumination (SLra) of 2537 and the long-wave light used in previous experiments. In Table V the record is divided into four consecutive periods. Remembering that the I+II end of the tube is the one toward the source of 2537, it will be noted that at first the fly went with increasing faithfulness to that end and then fell off to a considerable extent. This may indicate that the fly's eyes were at first more fatigued to the long waves, giving 2537 a greater effectiveness, and then became fatigued to 2537, more nearly equalizing the two. Possibly, even, long exposure to 2537 was destroying some receptive element in the fly's eyes.

Table VI summarizes a series of experiments, in each of which a fly bred in darkness was tested for 200 seconds in SLra, then subjected to either Lr or Sra for five minutes and again tested in SLra.

Let us consider first the five flies that were given Lr. Three of the five went less to the V end of the tube or, putting it the other way, more to the I end after they were subjected to the Lr than before. The opposite was true of the other two, but, on the whole there was a slight, but possibly significant shift in favor of Sra as though the five minutes in Lr had fatigued the flies to the long waves.

Considering the five flies that were given Sra, three showed a decided shift in favor of Lr. The other two showed no significant shift either way. On the whole, what may have been fatiguing to definite wavelengths seems to have been more marked in the case of Sra than in that of Lr.

The results of these experiments with light-fatigue are certainly not clear-cut enough to be very satisfactory. Possibly the fatiguing periods were not long enough, especially in view of the fact that for considerable portions of these periods the flies were not facing the light and that, in the case of the 2537 beam, the flies could get entirely out of it. However, while urging the desirability of further work on this problem, we do feel that the indications are that the 2537Å radiation has a physiological effect on the fly's eyes distinct from that of long light waves, just as the physiological effect on our eyes of blue is distinct from that of red, suggesting a distinct receptive mechanism. In other words, normal Drosophila melanogaster can see 2537Å radiation directly and was not reacting to an indirect effect of it. If this be true, it has some interesting bearings on biological speculation in view of the fact that light of so short a wave-length does not and probably never did occur in the environment of insects.

Table I.—Data from main experiments.

			ĎΪ					Dra				Lr	ľ				Sra					SLra	ಕ್ಷ	
	I	п	III	IV	>		П	H	IV	<b>&gt;</b>	I	11 11		Λ	<u> </u>	П	III	VI I	<u> </u>	Ι	П			<b>&gt;</b>
	146	154	108	83	110	65	63	67	   &	55		37 2	28 5	52 129	9 171	<u> </u>							42	90
	71	87	128	107	147	52	28	36	84	20	58	$\frac{30}{2}$	28 3	36   148		7 23	3 28	35	47		20	29		113
	125	75	111	81	208	40	49	50	45 1	911	39	29 7	73 3	31  128	8 180						23	24		125
	162	66	103	86	138	77	52	51	43	<u>.</u>		$\frac{35}{2}$	27 1	16 143	3 135		39	39				33		94
	150	87	06	88	185	22	22	47	30	91	55	$\frac{29}{2}$	27 3	32 157	7 140			30	49		40	29		99
	84	95	172	128	121	29	89	57	20	28		40	42 4	43 125	$5 103^{3}$	$3^3 45^3$	443	393			44	53		79
18;4 ♀	135	86	110	118	139	47	34	38	59 1	122	71 4	44 5		42 9						123	44	42	37	54
	123	97	107	112	161	52	45	54	26	63		37 5	51 4	7 111	1 184	38	3 28	3 26	24			33		8
	139	98	107	87	181	64	33	40		911	74	40 4	44 3	34 10			3 19	18		64	34	31	29	142
	141	85	128	103	143	51	22	65	99	91	46	26 3					37							79
	84	129	185	120	85	24	53 1	15	63	45	32	$\frac{28}{3}$	35 4	47   15	8  125	37	7 42	2 47	49	73	42	33		107
	152	101	104	121	122	22	42	47	47	68	43	17 3	31 3	32 17	7 201	1 36	3 22	24	. 17	_	33	27	20	115
	158	6	122	108	122	28	35	45	45 1	117	81	36 3	$\frac{31}{3}$	31 121	$\frac{1}{130}$	27	$\frac{1}{29}$	43	7	109	33	37		91
	182	98	104	84	144	74	20	44	42		23	59 3	35 4	44 139	9 115	5 42	34	1 28	81	121	33	36		71
26;2 ♀	129	64	128	113	166	06	44	64	44	28	<del>1</del> 60	30	32 3	38 136	6 131	11 51	88	29	51	134	<u> </u>	31	24	61
Total	1981	1433	1807	${1550}$		911	710 832 759	32 7.		1288 83	823 51	517 565	5 563	3 2032	2 2374	4 546	3 496	3 437	647	1487	610	513	523	1367

<sup>1</sup>Fly had trouble in walking.
<sup>2</sup>Only 9 minthes of Dr recorded.
<sup>3</sup>In one of the one-minute runs with Sra this fly "rested" for 34 seconds in the V section. Omitting this minute, the other four make the score: I, 98; III, 44; III, 31; IV, 32; V, 35.

Table II.—Data, reduced to percentages, showing the reaction to "visible" light

		Lr			$\mathbf{Dr}$			Lr-Dr	
	I, II	III	IV, V	I, II	III	IV, V	I, II	III	IV, V
 15;1, ♂	30	9	60	50	18	32		_ 9	28
15;2, ♂	29	9	61	29	24	47	0	—15	14
17;1, ♂	23	24	53	33	19	48	-10	5	5
18;1, ♀	38	9	53	44	17	39	<b>—</b> 6	- 8	14
18;2, ♀	28	9	63	40	15	46	-12	<b>— 6</b>	17
18;3, ♀	30	14	56	30	29	42	0	-15	14
18;4, ♀	38	17	45	39	18	43	<b>—</b> 1	<b>—</b> 1	2
18;5, ♀	30	17	53	37	18	46	<b>— 7</b>	<b>— 1</b>	7
19;1, ♂	38	15	47	38	18	45	0	- 3	2
19;2, ♂	24	10	66	38	21	41	-14	11	25
19;3, ♂	20	12	68	36	31	34	16	19	34
<b>24;2,</b> ♂	20	10	70	42	17	41	-22	<b>— 7</b>	29
<b>24;3</b> , ♂	39	10	51	41	20	38	_ 2	10	13
<b>24;4</b> , ♂	27	12	61	44	17	38	-17	_ 5	23
<b>26;2,</b> ♀	31	11	58	32	21	47	— 1	—10	11
Averages	30	12	58	38	20	42	8	-8	16

Table III.—Data, reduced to percentages, showing the reaction to 2537Å.

		Sra			Dra			Sra-Dra	L ,
	I, II	III	IV, V	I, II	III	IV, V	I, II	III	IV, V
15;1, ♂	71	15	14	43	26	31	28	11	-17
15;2, ♂	63	9	27	37	12	52	26	- 3	-25
17;1, ♂	69	9	22	30	17	54	39	- 8	-32
18;1, ♀	64	13	23	43	17	40	21	_ 4	-17
18;2, ♀	59	14	26	44	16	40	15	_ 2	-14
18;3, ♀¹	49	15	36	45	19	36	4	<b>— 4</b>	0
18;4, ♀	81	7	12	27	13	60	54	<b>—</b> 6	<b>—48</b>
18;5, ♀	74	9	17	32	18	50	42	<b>— 9</b>	-33
19;1, ♂	78	6	16	32	13	54	46	<b>— 7</b>	<b>—38</b>
19;2, ♂	66	12	21	26	22	52	40	-10	<b>—31</b>
19;3, ♂	54	14	32	26	38	36	28	24	<b>— 4</b>
24;2, ♂	79	7	14	39	16	45	40	- 9	-31
24;3, ♂	52	10	38	31	15	54	21	<b>— 5</b>	-16
<b>24;4</b> , ♂	52	11	36	41	15	44	11	<b>— 4</b>	<b>  -</b> 8
26;2, ♀	61	13	27	45	21	34	16	<b>—</b> 8	-7
Averages	65	11	24	36	19	45	29	-8	-21

<sup>1</sup>See footnote to Table 1. The revised scores would give the proportions in Sra, 59, 13, 28, and make Sra-Dra 14. This probably more nearly represents the reaction of the fly.

Table IV.—Data, reduced to percentages, showing the reaction when "visible" was opposed to 2537Å light.

		SLra			SLra–Dra	,
	I, II	III	IV, V	I, II	III	IV, V
15;1, ♂	46	10	44	3	—16	13
<b>15</b> ;2, ♂	41	10	49	4	_ 2	— 3
17;1, ♂	42	8	50	12	— 9	- 4
18;1, ♀	43	11	46	0	- 6	6
18;2, ♀	49	10	41	5	<b>—</b> 6	1
18;3, ♀	43	18	39	_ 2	1	3
18;4, ♀	. 56	14	30	29	1	30
18;5, ♀	53	11	36	21	<b>— 7</b>	-14
19;1, ♂	33	10	57	1	$-3 \\ -9$	3
19;2, ♂	. 48	13	39	22	9	- 3
19;3, ♂	38	13	49	12	-25	13
24;2, ♂	46	9	45	7	<b>— 7</b>	0
24;3, ♂	47	12	41	16	— 3	13
24;4, $\sigma$	53	12	35	12	<b>— 3</b>	<b>— 9</b>
<b>2</b> 6;2, ♀	61	10	28	16	—11	6
Averages	47	11	42	11	- 8	3

Table V.—Data from the experiment in which a fly (20;1) was kept in opposing lights for 2400 seconds.

	I+II	III	IV+V	I+II/IV+V
First Period	276	77	247	1.12
Second Period	301	63	236	1.28
Third Period	375	57	168	2.23
Fourth Period	309	49	242	1.28

Table VI.—Data from the attempts to induce differential fatigue of the flies' eyes.

	SLra ( followi	(200 se ng dar		follow	(200 se ing 5 m of Lr)	1		cond Sl ius the	
	I, II	III	IV, V	I, II	III	IV, V	I, II	III	IV, V
23;1, ♂	96	21	83	116	19	65	20	2	-18
23;3, ♀	101	30	69	110	21	69	9	— 9	0
30;1, ♀	85	28	87	85	17	98	0	11	11
30;4, ♀	75	26	99	89	26	85	14	0	14
31;1, ♀	74	23	103	75	30	95	1	7	<b>—</b> 8
Total	431	128	441	475	113	412	44	—15	<b>—29</b>
	1	(200 seing dar		1	(200 sec ing 5 m of Sra)	inutes	i .	cond SI ius the i	
	I, II	III	IV, V	I, II	III	IV, V	I, II	III	IV, V
23;2, ♂	82	23	95	80	26	94	2	3	— 1
29;1, ♂	95	24	81	49	16	135	46	- 8	54
30;2, ♂	83	33	84	75	20	105	- 8	-13	21
30;3, ♂	92	28	80	74	31	95	18	3	15
31;2, ♂	70	22	108	75	19	106	5	- 3	<b>—</b> 2
Total	422	130	448	353	112	535	69	18	87